

WE CLAIM:

1. A protein array comprising a surface having a plurality of spatially defined locations wherein at each location there are deposited at least two protein moieties which are capable of forming a complex characterised in that said complex is transiently formed.

2. The protein array of claim 1 wherein the complex is transiently formed during catalysis.

3. A protein array comprising a surface having a plurality of spatially defined locations wherein at each location there are deposited at least two protein moieties characterised in that said protein moieties at each location act sequentially on a substrate of interest.

4. The protein array of any one of claims 1 to 3 wherein at least one of said protein moieties at each location is capable of being membrane-associated or membrane-bound or has been modified to interact with a non-polar or amphipathic molecule.

5. The protein array of any one of claims 1 to 4 wherein at least one of said moieties at each location is a drug metabolising enzyme.

6. A protein array comprising a surface upon which are deposited at spatially defined locations at least two protein moieties characterised in that said protein moieties are derived from one or more drug metabolising enzymes.

7. The protein array of claim 5 or claim 6 wherein at least one of said protein moieties at each location is a P450 protein.

8. The protein array of claim 3 or claim 6 wherein said protein moieties are attached to said surface through a marker moiety appended to each protein moiety.
9. The protein array of any one of claims 1 to 7 wherein said protein moieties are incorporated into a membrane, vesicle or liposome which is immobilised in proximity to said surface.
10. The protein array of claim 5 or claim 6 wherein said drug metabolising enzymes are selected from the group consisting of cytochrome P450s, flavin monooxygenases, UDP-glycosyltransferases, glutathione S-transferases, sulfotransferases and N-acetyltransferases.
11. The protein array of any one of claims 3 to 10 wherein one or more Phase 1 drug metabolising enzymes and one or more Phase 2 drug metabolising enzymes are present on the array.
12. The protein array of any one of claims 5 to 10 wherein said drug metabolising enzymes are *H. sapiens* cytochrome P450s and are selected from the group consisting of CYP1A2, CYP2A6, CYP2B6, CYP2C8, CYP2C9*1, CYP2C9*2, CYP2C9*3, CYP2C19, CYP2D6, CYP2E1, CYP3A4, CYP3A5.
13. The protein array of any one of claims 5 to 12 wherein one or more of said drug metabolising enzymes are derived from different ethnic groups, different genders, different mammalian species, or different mutant versions of a wild type enzyme.
14. A method of making a protein array comprising the steps of:
- providing two or more drug metabolising enzymes of interest from either recombinant, native or synthetic sources;
 - depositing said proteins at spatially defined locations on a surface to give an array.

15 The method of claim 14, wherein said drug metabolising enzymes are brought
into contact with the array in admixture with other protein molecules and deposition
on the array occurs with simultaneous purification of the protein moiety on the array
5 via a tag incorporated in the protein moiety.

16. The method of claim 14 or claim 15, wherein said drug metabolising enzymes
are deposited with other proteins from an expression host cell on a surface at spatially
defined locations to give an array.

10 17. A method of making a protein array comprising the steps of:

a) providing one or more proteins from either recombinant, native or synthetic
sources incorporated in purified or partially purified membrane or membrane-like
preparations;

15 b) arraying said proteins by encapsulation of said membrane or membrane-like
preparations into a gel matrix which is deposited on the surface.

18. A method of making an array of drug metabolising enzymes comprising the
steps of:

20 a) providing drug metabolising enzymes from either recombinant, native or
synthetic sources in the form of purified or partially purified membrane or membrane-
like preparations;

b) arraying said drug metabolising enzymes either by deposition of said
membrane or membrane-like preparations onto a suitable surface capable of capturing
25 the membranes or by encapsulation of said membrane or membrane-like preparations
into a gel matrix which is deposited on the surface.

19. The method of claim 17 or 18 wherein one or more of said membrane or
membrane-like preparations contains two or more different proteins.

20. The method of claim 19 wherein said two or more different proteins are capable of forming a complex with each other.
21. The method of claim 20 wherein said complex is transiently formed.
22. The method of claim 19 wherein said two or more different proteins act sequentially on a substrate of interest.
23. An array made by the method of any one of claims 14 to 22.
24. A method of screening a set of protein moieties for molecules which interact with one or more proteins comprising the steps of
- a) bringing one or more test molecules into contact with an array as claimed in any one of claims 1 to 13 or 23; which carries said set of protein moieties;
 - b) detecting an interaction between one or more test molecules and one or more proteins on the array.
25. A method of simultaneously determining the relative properties of members of a set of protein moieties, comprising the steps of:
- a) bringing an array as claimed in any one of claims 1 to 13 or 23 which carries said set of protein moieties into contact with one or more test substances, and
 - b) observing the interaction of said test substances with the set members on the array.
26. The method of claim 25 wherein one or more of said protein moieties are drug metabolising enzymes and wherein said enzymes are activated by contact with an accessory protein or by chemical treatment.
27. Use of an array as claimed in any one of claims 1 to 13 or 23 in the examination of gender differences in drug metabolism.

28. Use of an array as claimed in any one of claims 1 to 13 or 23, in the examination of ethnicity-related differences in drug metabolism and toxicity.
29. Use of an array as claimed in any one of claims 1 to 13 or 23 in the examination of differences in drug metabolism between two or more mammalian species.
30. The use as defined in claim 29 wherein said mammalian species are human and rat.
31. Use of an array as claimed in any one of claims 1 to 13 or 23 in the examination of the cytotoxicity of drug metabolites.
32. Use of an array as claimed in any one of claims 1 to 13 or 23 in the definition and quantitation of metabolic pathways for small molecules.
33. Use of an array as claimed in any one of claims 1 to 13 or 23 in the screening of compounds for selectivity in their ability to bind and inhibit individual drug metabolising enzymes.
34. Use of an array as claimed in any one of claims 1 to 13 or 23 in the analysis of the induction of P450 expression by one or more compounds of interest.
35. Use of an array as claimed in any one of claims 1 to 13 or 23 in the analysis of the effects of mutation on the activity of a drug metabolising enzyme of interest.
36. A method of expressing and purifying a drug metabolising enzyme (DME), comprising the steps of:
- expressing a DME of interest in a host cell;
 - subjecting said host cell to conditions suitable to lyse the cell;
 - obtaining a membrane associated cell fraction from the lysed cell;

- d) solubilising said membrane associated cell fraction by the addition of a detergent;
- e) after an incubation period sufficient to solubilise the DME protein contained in said membrane associated cell fraction, performing a further centrifugation step to
5 produce a supernatant containing said DME protein;
- f) subjecting said supernatant to chromatography to purify said DME protein.